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Comparative Evaluation of Microhardness of Two Different Remineralizing Pastes - An In vitro study

Research Article

DR. P. Niharika[1] ,Dr. Subash Sharma[2*]

[1] Department of Conservative Dentistry and Endodontics, Saveetha Dental College and Hospital, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai, India
[2] Ass.Professor, Head of the Department, Department of Aesthetic dentistry, Saveetha Dental college and Hospital, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai, IndiaSaveetha University, Chennai-77, India.

Abstract

Aim : The aim of this study is to evaluate the remineralization potential of two different remineralizing agents.
Objective: To evaluate the remineralizing potential with two different remineralizing agents. To assess the microhardness before and after application of remineralising agents.
Materials And Methods: Total of 16 samples were taken divided in 3 groups: Group 1 – GC tooth mousse Group 2 – Ena-Fix dentifrice group 3 – Colgate total. Pre microhardness testing was done using Vickers hardness testing machine, remineralization was carried out with group A, B, C using a brushing simulator. Post microhardness testing was done to evaluate the remineralization potential among three different remineralizing agents.
Results: Comparing the two groups at baseline, post cycle the statistical analysis was done using paired t test, Anova . Group A – GC tooth mousse with p > 0.096, Group B – EnaFix P > 0.542 Group C – P >0.003 which had shown significant difference between all three groups
Conclusion: The use of CPP-ACP and EnaFix in recent years have been possible methods in prevention of enamel caries and in halting the progress of the existing enamel lesion. CPP -ACP containing remineralizing paste has shown better remineralizing effect followed by EnaFix and Colgate total.

Keywords: Remineralizing Agents; Casein Phosphate; Amorphous Calcium Phosphate; Enamel Caries.

Introduction

Dental caries is a process that takes place on any tooth surface where the dental plaque is accumulated for a long period. Initial caries progression may be prevented by suitable surface treatment[1]. This issue signifies the current concept regarding remineralization and demineralization of the tooth surface[2].

The first step in the remineralization process is demineralization, which regulates and reverses the decay process. Demineralization happens as acidogenic bacteria reduce the pH of the calculus. The remineralization process starts when Ca+2 and PO4 ions in saliva raise the pH of calculus. Therefore, demineralized lesions are remineralized. However, when the demineralization is equal to or higher than remineralization, decay occurs [3].

The level of Ca^{+2} and PO4 in the saliva has a big impact on the buffer ability of the saliva. When fluoride ions are present in the saliva, the rate of remineralization increases. As a result, research focused on the role of fluoride in preventing caries and reversing the decay or demineralization phase. In recent in vivo and in vitro studies, the effect of fluoride on remineralization and demineralization has been researched [4]

One of the most effective remineralizing agents for caries prevention is fluoride. Nonetheless, there have been several concerns posed about fluorosis and fluoride intake in general. Fluoride replacements such as CPP-ACP and nano-hydroxyapatite (NHA) have been suggested in recent years due to their anticariogenic properties.[5,6]. Earlier studies have shown that the application of a CPPs toothpaste and sodium fluoride (Colgate Neutrafluor 9,000 ppm) (NaF) can provide significant additional prevention of enamel demineralisation when resin-modified glass ionomer cement (RMGIC) is used for bonding molar tubes for orthodon-

***Corresponding Author:**
Dr. Subash Sharma,
Ass.Professor, Head of the Department, Department of Aesthetic dentistry, Saveetha Dental college and Hospital, Saveetha Institute of Medical and Technical Sciences, Saveetha University, Chennai, India
Tel : +919884533118
E-mail : subash@saveetha.com

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tic patient as preventive actions[7].

An in vitro study to evaluate the remineralisation of incipient enamel lesions by the topical application of Casein Phosphopeptide-Amorphous Calcium Phosphate (CPP-ACP) using laser fluorescence and scanning electron microscope showed high scores of remineralisation[8] .

Micro hardness and roughness are two of the important properties of materials and associated with loss or gain of mineral content in tooth structure[9]. An ideal toothpaste should remove unwanted surface deposits and stains with minimal influence on the enamel, dentine and restorations [10]. Thus, the effects of such products on properties of enamel and restorative materials are important [11]. It is one of the important properties of the materials which correlates with strength, proportional limit and wear resistance [12].

Therefore, due to a continuous (daily) therapeutic recommendation for using these products, it is relevant to assess the effects of these toothpastes on properties such as microhardness and its remineralisation property. This is an in-vitro study that aims at evaluation of the microhardness and the remineralising property of 3 different remineralising toothpastes.Previously our team has a rich experience in working on various research projects across multiple disciplines (Govindaraju and Gurunathan 2017; A. Christabel et al. 2016; Soh and Narayanan 2013; Mehta et al. 2019; Ezhilarasan, Apoorva, and Ashok Vardhan 2019; Campeau et al. 2014; Kumar and S 2016; S. L. Christabel 2015; Kumar and Rahman 2017; Sridharan, Ramani, and Patankar 2017; Ramesh et al. 2016; Thamaraiselvan et al. 2015; Thangaraj et al. 2016; Ponnulakshmi et al. 2019; "Fluoride, Fluoridated Toothpaste Efficacy and Its Safety in Children - Review" 2018) Now the growing trend in this area motivated us to pursue this project.

Materials And Methods

It was an in vitro study conducted in the month of November 2020 – February 2021 in the city of Chennai, Tamil Nadu.

Ethical Approval

The study was registered with the Institutional Review Board of the Saveetha Institute of Medical and Technical Sciences, Chennai, Tamil Nadu, India. Ethical approval was obtained from the Institutional Review Board of the SIMATS.

Sampling Method

Sixteen freshly extracted sound teeth, extracted for therapeutic reasons, were used. Teeth with any caries or white spot lesions were excluded. The teeth were decoronated and the crown portions were divided into two segments of one buccal and one palatal half using a diamond disc mounted on a contra-angle handpiece. Enamel samples were fixed on the self-cure acrylic mold with the enamel surface exposed. These samples were stored in deionized water until further use.

Group A: GC Tooth Mousse
Group B: Enafix
Group C: Colgate Total

Remineralisation was carried out using the brushing stimulator. After multiple cycles of remineralization, the surface microhardness of the specimens was determined using Vickers microhardness testing machine

Statistical Analyses

The collected data was tabulated into Microsoft office Excel 2013 transferred to SPSS version 26.0 software (SPSS, Chicago, IL, USA) for statistical analysis. This data was analyzed statistically using the paired t test to evaluate differences in the values obtained pre and post cycle and to see if the results were statistically significant. The baseline data was evaluated and compared against the post cycle values for all the 3 remineralising toothpastes. The confidence interval was set at 95%.

Results

The result showed there has been a statistically significant difference in the microhardness among the three samples. Microhardness results per sample and treatment are shown below in Table 1. All tooth samples underwent statistically similar levels of demineralisation and remineralization. The various p values found for group A (0.096), group B (0.542), group C (0.003). None of the values were statistically significant.

Discussion

Hardness is a surface property of a material that shows its resistance against permanent deformation. Vickers hardness is a type of microhardness test which is commonly used to evaluate surface microhardness of brittle and restorative materials [12, 28–31].

Table 1 :The above table showing the baseline and post cycle values before and after microhardness testing for the total (16 samples) which were grouped as Group A , Group B , Group C . Mean , Standard deviation values were interpreted and Statistical analysis was done using Anova , t-test and Post -hoc tests were calculated and P value was interpreted .

Groups	MEAN		Standard deviation		t-test	P value
	Baseline	Post Cycle	Baseline	Post Cycle		
Group A (GC mousse)	329.550	366.167	2.654	71.474	-2.047	.096
Group B (Enafix Dentifrice)	365.033	368.833	-1.285	-40.141	-654	.542
Group C (Colgate Total)	482.900	335.000	1.835	8.514	8.514	0.003

Figure 1: Sample Preparation For Groups A,B And C. Mounted Samples On Die Stone.

Figure 2:Brushing Simulator

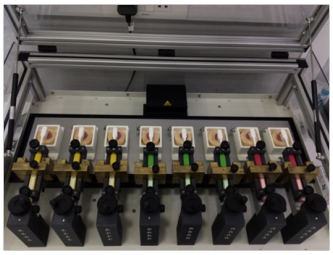


Figure 3:Vickers Hardness Tester

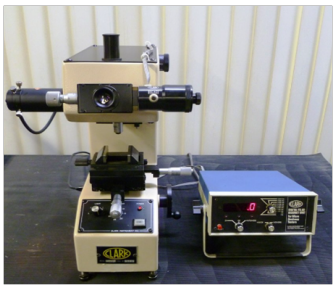
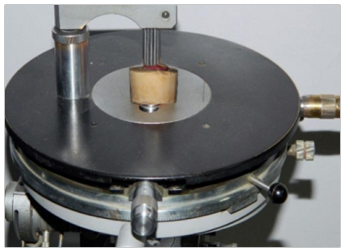


Figure 4:Vickers Hardness Tester (Measurement Of Sample)



Many commercially available ones contain ingredients that may have adverse effects on the surface of restorations and teeth[32]. Little information was available about the adverse effects of some of these new dentifrices [10]. Fluoridated toothpastes have been used extensively and their anti-cariogenic properties have been evaluated through laboratory, clinical and epidemiological studies[33]. The increased use of these toothpastes suggests that a more accurate assessment of their effectiveness is needed. Micro hardness determination can provide indirect evidence of demineralization and remineralization in dental hard tissues[34].

The goal of this project was to evaluate the in vivo effects of a novel mouthwash on enamel recovery after demineralization by simulating an artificial intra-oral exposure by using the brushing stimulator. The results showed significantly higher microhardness values following application of the toothpastes.

About the fact that GC tooth mousse supplies calcium and phosphate ions for remineralization, the microhardness was mild. According to Galbiatti et al., after CCP-ACP application, non-homogeneous deposits with adherent irregularities were developed and settled as globular structures .These globular structures occupy the interprismatic cavities and surround the prisms partly[35]. These findings were consistent with the study done by Ferrazzano, 2011[36].

A study by Newby[37] stated that 1150 ppm NaF test dentifrice and Crest Cavity Protection (1100 ppm NaF) protected enamel specimens greater than the fluoride-free placebo. In another study by HatipogluNaF toothpastes significantly increased the microhardness of the lesions ($p<0.001$) when compared to control groups and revealed a mineral precipitation band on the surface layer of all samples[38].

Haghgoo et al[39] (2014) compared between NHA and NaF mouthwashes and found no difference in the remineralizing effect. However, surface microhardness and tooth remineralization significantly increased. They used the remineralizing agent in the form of a mouthwash. In an in vivo analysis, Najibfard et al[40]

(2011) compared 10% and 5% NHA, as well as a mixture of 10% NHA + 1100 ppm NAF. The findings of the microradiographs revealed that the toothpastes tested had similar remineralizing effects.

Our findings revealed that toothpaste from group C has a greater remineralizing effect. However, by using the Vickers microhardness tester to measure the remineralizing effect of agents, researchers should be mindful of the method's drawbacks and the limitations of extrapolating in vitro effects to the clinical environment. This procedure is not capable of completely simulating oral conditions.

Our institution is passionate about high quality evidence based research and has excelled in various fields (JayaseelanVijayashreePriyadharsini 2019; Pc, Marimuthu, and Devadoss 2018; Ramesh et al. 2018; Ramadurai et al. 2019; Sridharan et al. 2019;Ezhilarasan, Apoorva, and Ashok Vardhan 2019; Mathew et al. 2020; Samuel 2021; R et al. 2020; Chandrasekar et al. 2020; J. VijayashreePriyadharsini, SmilineGirija, and Paramasivam 2018)

Conclusion

All the three toothpastes showed remineralising potential and there was an increase in the overall microhardness but the values were not statistically significant. Future research should concentrate on assessing the possible impacts of synthetic CPPs in order to develop a new prevention approach focusing on the use of these bio-active concepts in personal hygiene products to mitigate cariogenicity.

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References

[1]. Fejerskov O, Kidd E. Dental Caries: The Disease and Its Clinical Management. John Wiley & Sons; 2009.
[2]. Hoceini A, Khelil NK, Ben-Yelles I, Mesli A, Ziouani S, Ghellai L, et al. Caries-related factors and bacterial composition of supragingival plaques in caries free and caries active Algerian adults. Asian Pacific journal of tropical biomedicine. 2016 Aug 1;6(8):720-6.
[3]. Peters MC, Bresciani E, Barata TJ, Fagundes TC, Navarro RL, Navarro MF, et al. In vivo dentin remineralization by calcium-phosphate cement. J Dent Res. 2010 Mar;89(3):286-91. Pubmed PMID: 20139340.
[4]. Schemehorn BR, Orban JC, Wood GD, Fischer GM, Winston AE. Remineralization by fluoride enhanced with calcium and phosphate ingredients. J Clin Dent. 1999;10(1 Spec No):13-6. Pubmed PMID: 10686853.
[5]. Vyavhare S, Sharma DS, Kulkarni VK. Effect of three different pastes on remineralization of initial enamel lesion: an in vitro study. J Clin Pediatr Dent. 2015 Winter;39(2):149-60. Pubmed PMID: 25823485.
[6]. Kalra DD, Kalra RD, Kini PV, Prabhu CA. Nonfluoride remineralization: An evidence-based review of contemporary technologies. Journal of Dental and Allied Sciences. 2014 Jan 1;3(1):24.
[7]. Shivananda DH, Ansar W, Dinsha AR, Sam G, Bharath S, Kakti A, et al. Effectiveness of Various Dental Varnishes in Prevention of Enamel Demineralization around Orthodontic Brackets: An In Vitro Study. J Contemp Dent Pract. 2020 Jun 1;21(6):621-625. Pubmed PMID: 33025929.
[8]. Pai D, Bhat SS, Taranath A, Sargod S, Pai VM. Use of laser fluorescence and scanning electron microscope to evaluate remineralization of incipient enamel lesions remineralized by topical application of casein phospho peptide amorphous calcium phosphate (CPP-aCP) containing cream. J Clin Pediatr Dent. 2008 Spring;32(3):201-6. Pubmed PMID: 18524269.
[9]. Joiner A. Review of the effects of peroxide on enamel and dentine properties. J Dent. 2007 Dec;35(12):889-96. Pubmed PMID: 17964705.
[10]. Watanabe MM, Rodrigues JA, Marchi GM, Ambrosano GM. In vitro cariostatic effect of whitening toothpastes in human dental enamel-microhardness evaluation. Quintessence Int. 2005 Jun;36(6):467-73. Pubmed PMID: 15954253.
[11]. Wattanapayungkul P, Yap AU, Chooi KW, Lee MF, Selamat RS, Zhou RD. The effect of home bleaching agents on the surface roughness of tooth-colored restoratives with time. Oper Dent. 2004 Jul-Aug;29(4):398-403. Pubmed PMID: 15279478.
[12]. Craig RG, Sakaguchi RL. Powers JM. Restorative dental materials. St. Louis: Mosby. 2002.
[13]. Govindaraju L, Gurunathan D. Effectiveness of Chewable Tooth Brush in Children-A Prospective Clinical Study. J Clin Diagn Res. 2017 Mar;11(3):ZC31-ZC34. Pubmed PMID: 28511505.
[14]. Christabel A, Anantanarayanan P, Subash P, Soh CL, Ramanathan M, Muthusekhar MR, et al. Comparison of pterygomaxillary dysjunction with tuberosity separation in isolated Le Fort I osteotomies: a prospective, multi-centre, triple-blind, randomized controlled trial. Int J Oral Maxillofac Surg. 2016 Feb;45(2):180-5. Pubmed PMID: 26338075.
[15]. Soh CL, Narayanan V. Quality of life assessment in patients with dentofacial deformity undergoing orthognathic surgery--a systematic review. Int J Oral Maxillofac Surg. 2013 Aug;42(8):974-80. Pubmed PMID: 23702370.
[16]. Mehta M, Deeksha, Tewari D, Gupta G, Awasthi R, Singh H, et al. Oligonucleotide therapy: An emerging focus area for drug delivery in chronic inflammatory respiratory diseases. Chem Biol Interact. 2019 Aug 1;308:206-215. Pubmed PMID: 31136735.
[17]. Ezhilarasan D, Apoorva VS, Ashok Vardhan N. Syzygium cumini extract induced reactive oxygen species-mediated apoptosis in human oral squamous carcinoma cells. J Oral Pathol Med. 2019 Feb;48(2):115-121. Pubmed PMID: 30451321.
[18]. Campeau PM, Kasperaviciute D, Lu JT, Burrage LC, Kim C, Hori M, et al. The genetic basis of DOORS syndrome: an exome-sequencing study. Lancet Neurol. 2014 Jan;13(1):44-58. Pubmed PMID: 24291220.
[19]. Kumar S, Sneha S. Knowledge and awareness regarding antibiotic prophylaxis for infective endocarditis among undergraduate dental students. Asian Journal of Pharmaceutical and Clinical Research. 2016;154.
[20]. Christabel SL, Gurunathan D. Prevalence of type of frenal attachment and morphology of frenum in children, Chennai, Tamil Nadu. World J Dent. 2015 Oct;6(4):203-7.
[21]. Kumar S, Rahman RE. Knowledge, awareness, and practices regarding biomedical waste management among undergraduate dental students. Asian Journal of Pharmaceutical and Clinical Research. 2017;10(8):341.
[22]. Sridharan G, Ramani P, Patankar S. Serum metabolomics in oral leukoplakia and oral squamous cell carcinoma. J Cancer Res Ther. 2017 Jul-Sep;13(3):556-561. Pubmed PMID: 28862226.
[23]. Ramesh A, Varghese SS, Doraiswamy JN, Malaiappan S. Herbs as an antioxidant arsenal for periodontal diseases. J Intercult Ethnopharmacol. 2016 Jan 27;5(1):92-6. Pubmed PMID: 27069730.
[24]. Thamaraiselvan M, Elavarasu S, Thangakumaran S, Gadagi JS, Arthie T. Comparative clinical evaluation of coronally advanced flap with or without platelet rich fibrin membrane in the treatment of isolated gingival recession. J Indian Soc Periodontol. 2015 Jan-Feb;19(1):66-71. Pubmed PMID: 25810596.
[25]. Thangaraj SV, Shyamsundar V, Krishnamurthy A, Ramani P, Ganesan K, Muthuswami M, et al. Molecular Portrait of Oral Tongue Squamous Cell Carcinoma Shown by Integrative Meta-Analysis of Expression Profiles with Validations. PLoS One. 2016 Jun 9;11(6):e0156582. Pubmed PMID: 27280700.
[26]. Ponnulakshmi R, Shyamaladevi B, Vijayalakshmi P, Selvaraj J. In silico and in vivo analysis to identify the antidiabetic activity of beta sitosterol in adipose tissue of high fat diet and sucrose induced type-2 diabetic experimental rats. Toxicol Mech Methods. 2019 May;29(4):276-290. Pubmed PMID: 30461321.
[27]. Ramakrishnan M, Bhurki M. Fluoride, Fluoridated Toothpaste Efficacy And Its Safety In Children-Review. International Journal of Pharmaceutical Research. 2018 Oct 1;10(04):109-14.
[28]. Sasaki RT, Arcanjo AJ, Flório FM, Basting RT. Micromorphology and microhardness of enamel after treatment with home-use bleaching agents containing 10% carbamide peroxide and 7.5% hydrogen peroxide. J Appl Oral Sci. 2009 Nov-Dec;17(6):611-6. Pubmed PMID: 20027436.
[29]. Taher NM. The effect of bleaching agents on the surface hardness of tooth colored restorative materials. J Contemp Dent Pract. 2005 May 15;6(2):18-26. Pubmed PMID: 15915201.
[30]. Rodrigues JA, Marchi GM, Ambrosano GM, Heymann HO, Pimenta LA. Microhardness evaluation of in situ vital bleaching on human dental enamel using a novel study design. Dent Mater. 2005 Nov;21(11):1059-67. Pubmed PMID: 16143381.
[31]. Türker SB, Biskin T. The effect of bleaching agents on the microhardness of

dental aesthetic restorative materials. J Oral Rehabil. 2002 Jul;29(7):657-61. Pubmed PMID: 12153455.

[32]. Meyers IA, McQueen MJ, Harbrow D, Seymour GJ. The surface effect of dentifrices. Aust Dent J. 2000 Jun;45(2):118-24. Pubmed PMID: 10925508.

[33]. Rahardjo A, Karina K, Fadhilah A, Eriwati YK, Triaminingsih S, Maharani DA. Caries-şpreventive Effect of 1300ppm Fluoride and Carrageenan Containing Toothpaste. Journal of Dentistry Indonesia. 2013 Sep 22;20(1):1-ş4.

[34]. Unlü N, Cobankara FK, Altinöz C, Ozer F. Effect of home bleaching agents on the microhardness of human enamel and dentin. J Oral Rehabil. 2004 Jan;31(1):57-61. Pubmed PMID: 15125598.

[35]. Carvalho FG, Brasil VL, Silva Filho TJ, Carlo HL, Santos RL, Lima BA. Protective effect of calcium nanophosphate and CPP-ACP agents on enamel erosion. Braz Oral Res. 2013 Nov-Dec;27(6):463-70. Pubmed PMID: 24346043.

[36]. Ferrazzano GF, Amato I, Cantile T, Sangianantoni G, Ingenito A. In vivo remineralising effect of GC tooth mousse on early dental enamel lesions: SEM analysis. Int Dent J. 2011 Aug;61(4):210-6. Pubmed PMID: 21851353.

[37]. Newby CS, Creeth JE, Rees GD, Schemehorn BR. Surface microhardness changes, enamel fluoride uptake, and fluoride availability from commercial toothpastes. J Clin Dent. 2006;17(4):94-9. Pubmed PMID: 17131711.

[38]. HATİPOĞLU Z, YAVLAL GÖ, YAVLAL Ö. Effects of Different Fluoride-containing Toothpastes on In Vitro Enamel Remineralization. Bezmialem Science. 2019;7(1):12.

[39]. Haghgoo R, Rezvani MB, Salehi Zeinabadi M. Comparison of nano-hydroxyapatite and sodium fluoride mouthrinse for remineralization of incipient carious lesions. J Dent (Tehran). 2014 Jul;11(4):406-10. Pubmed PMID: 25584051.

[40]. Najibfard K, Ramalingam K, Chedjieu I, Amaechi BT. Remineralization of early caries by a nano-hydroxyapatite dentifrice. J Clin Dent. 2011;22(5):139-43. Pubmed PMID: 22403978.